WO 2005/034844

680	KMHD*R*LEPEDEESDADYDYQN*R*L*A*AALGIR	709	SEQ ID NO. 1
HC1	KMHDRLEPED		SEQ ID NO. 2
HC2	LEPEDEESDA		SEQ ID NO. 3
HC3	EESDADYDYQ		SEQ ID NO. 4
HC4	DYDYQNRLAA		SEQ ID NO. 5
HC5	NRLAAALGIR		SEQ ID NO. 6

FIG. 1

PCT/US2004/021487

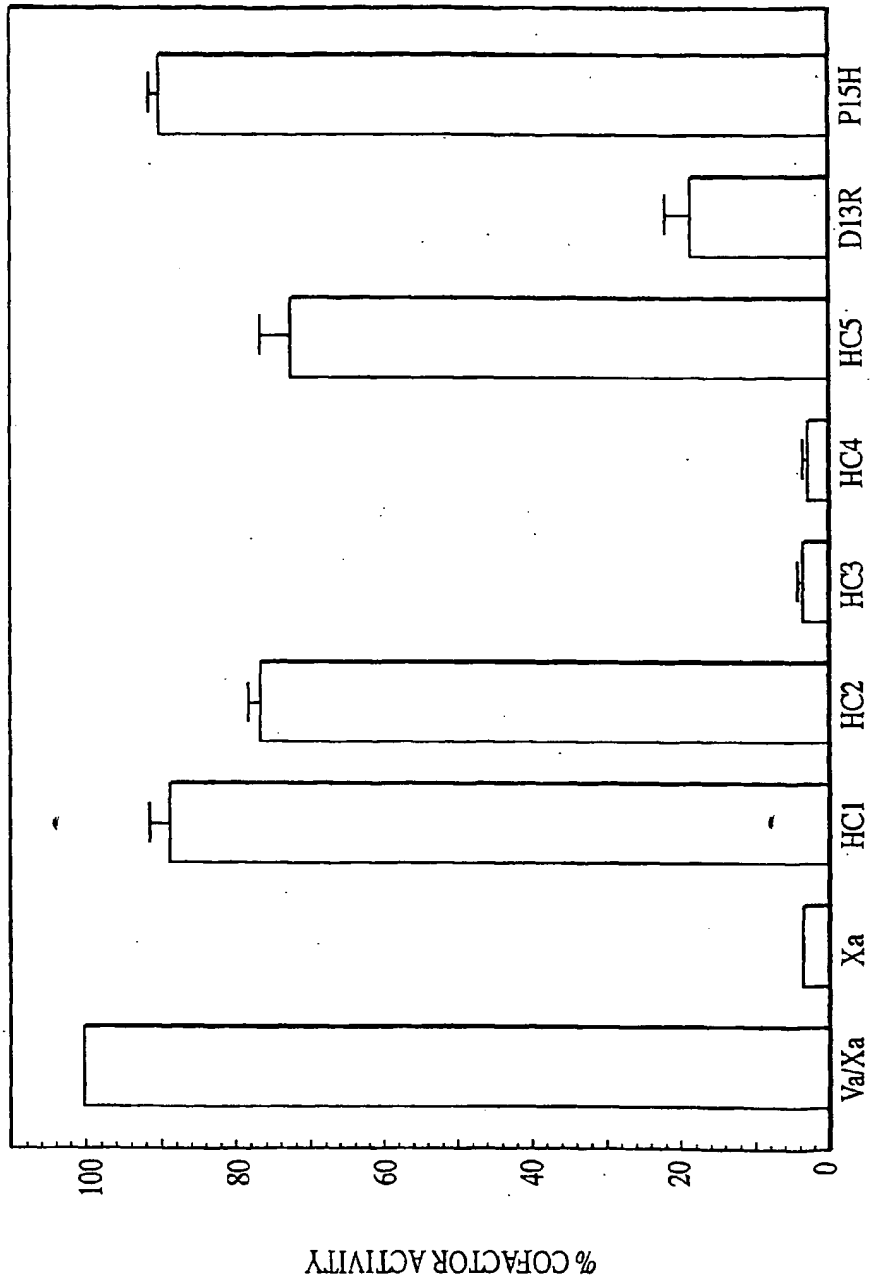
% COFACTOR ACTIVITY
0
20
40
60
80
100
Va/Xa
Xa
HC1
HC2
HC3
HC4
HC5
D13R
P15H

FIG. 2A

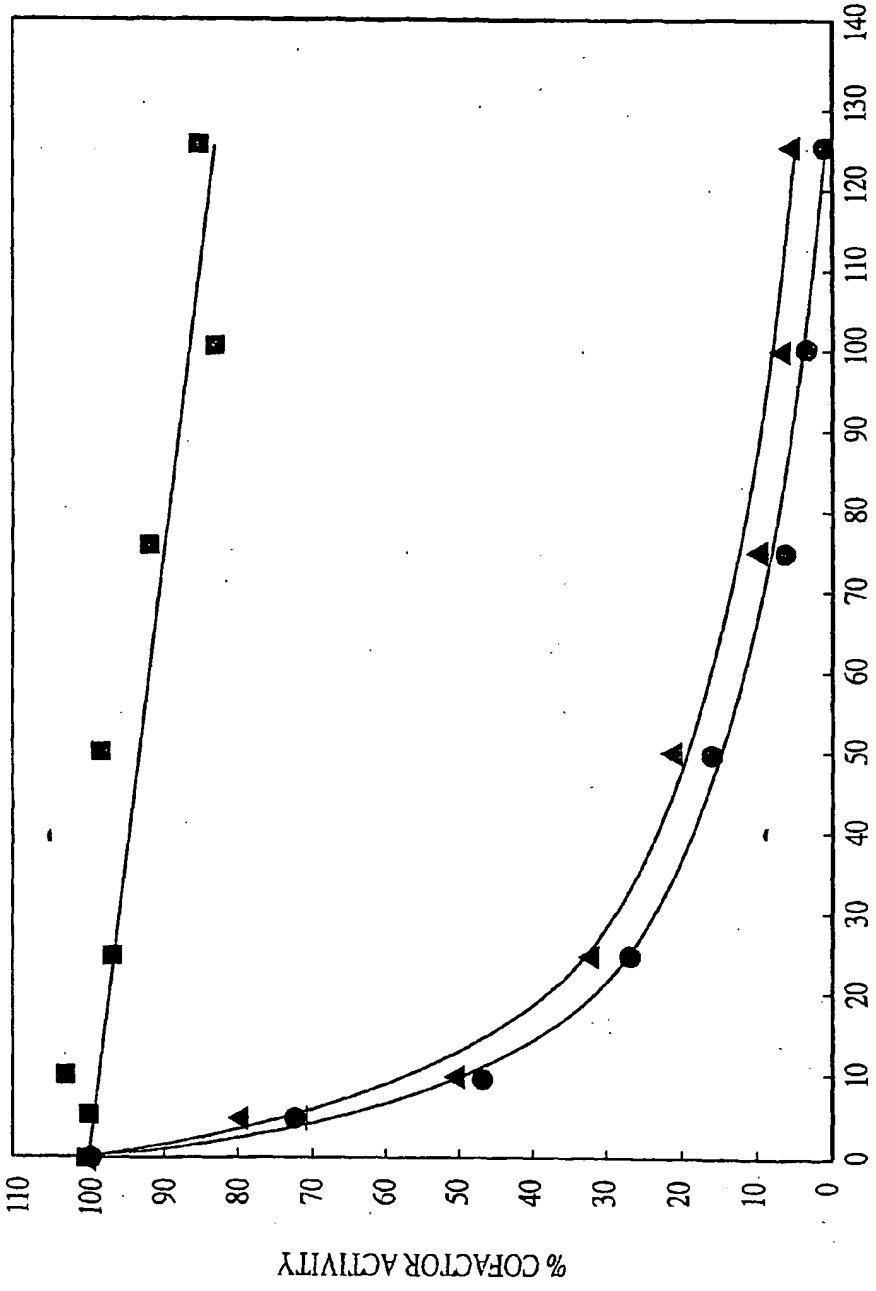
% COFACTOR ACTIVITY
110
100
90
80
70
60
50
40
30
20
10
0
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
PEPTIDE CONCENTRATION (μM)

FIG. 2B

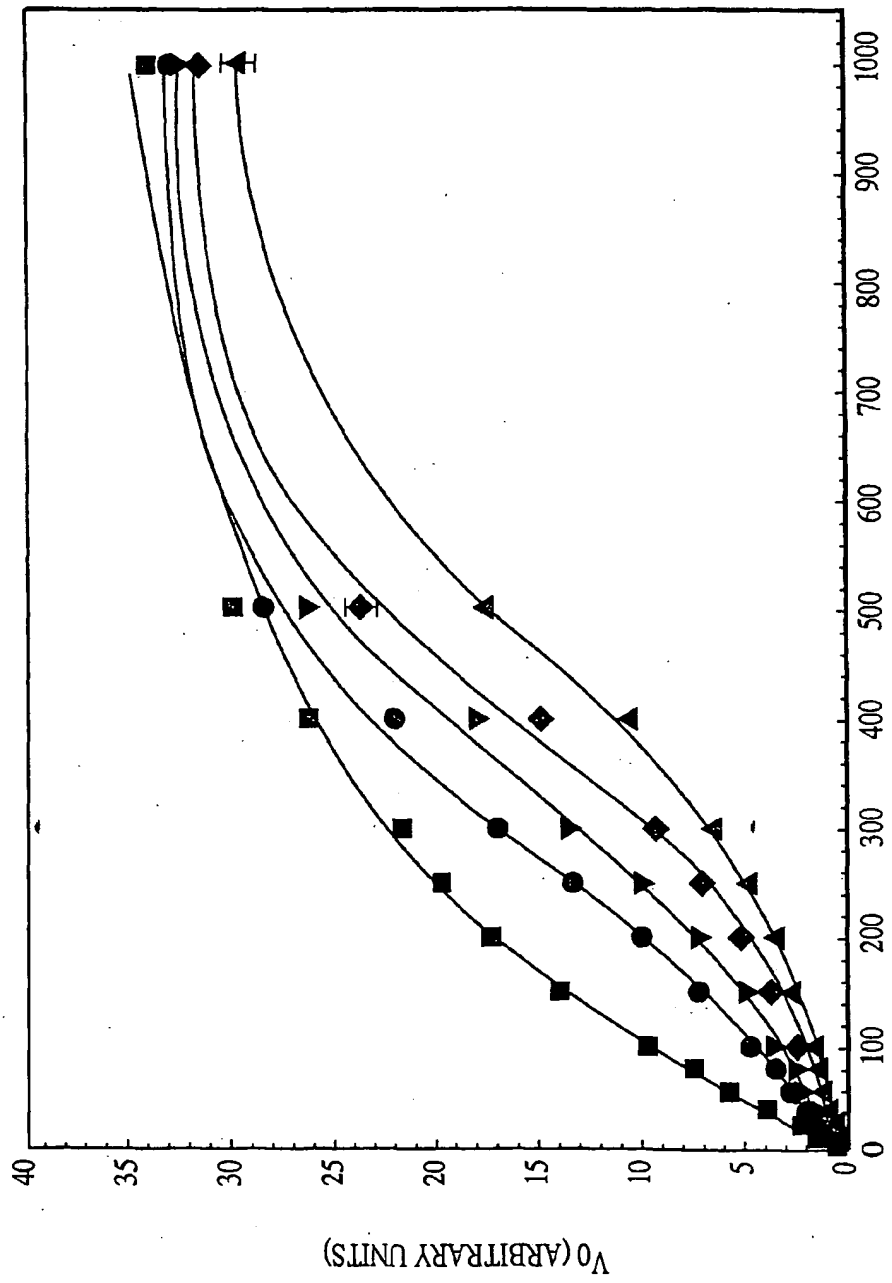
V_0 (ARBITRARY UNITS)
0
5
10
15
20
25
30
35
40
0
100
200
300
400
500
600
700
800
900
1000
PROTHROMBIN (nM)

FIG. 2C

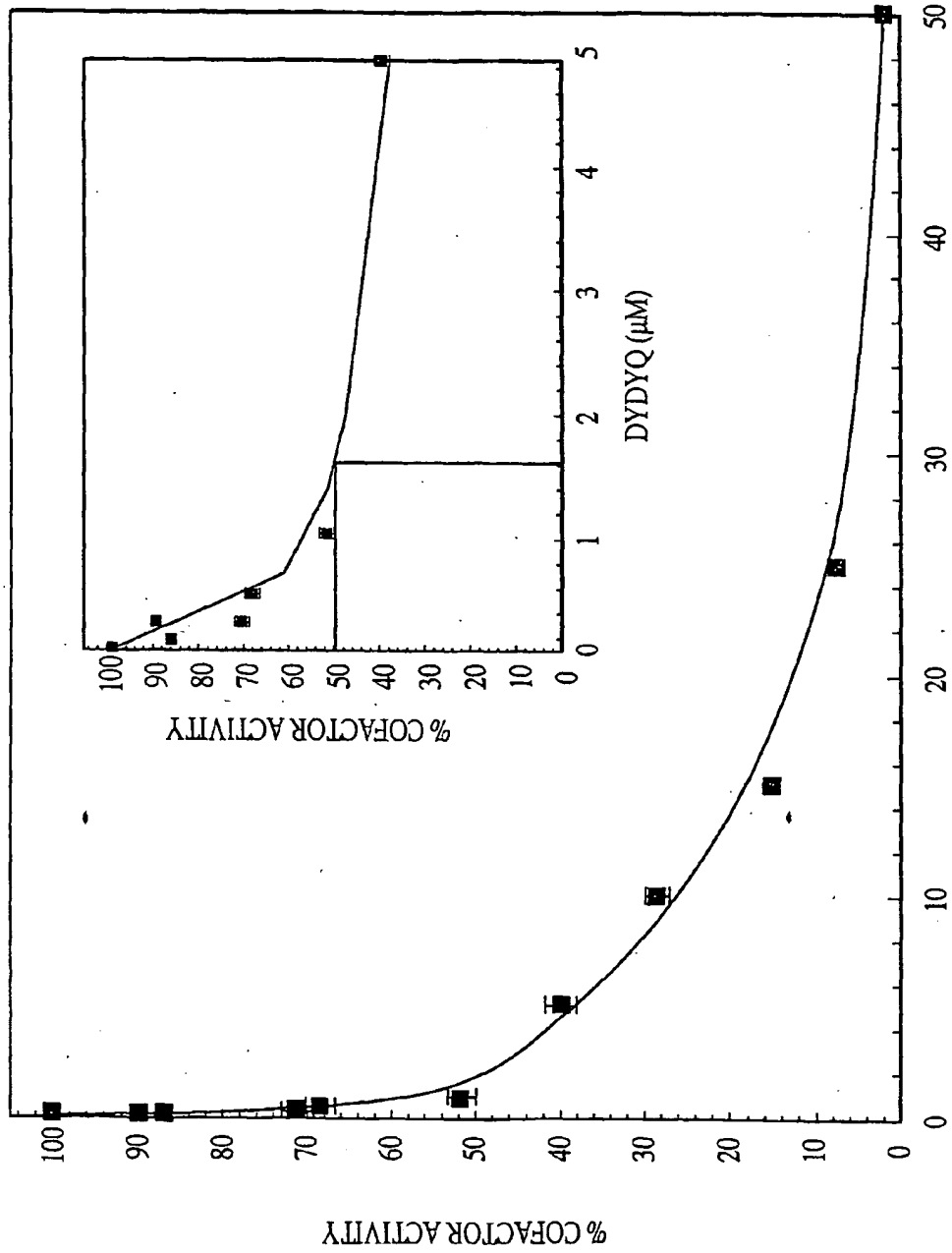
% COFACTOR ACTIVITY
PEPTIDE CONCENTRATION (μM)
DYDYQ (μM)
% COFACTOR ACTIVITY

FIG. 3A

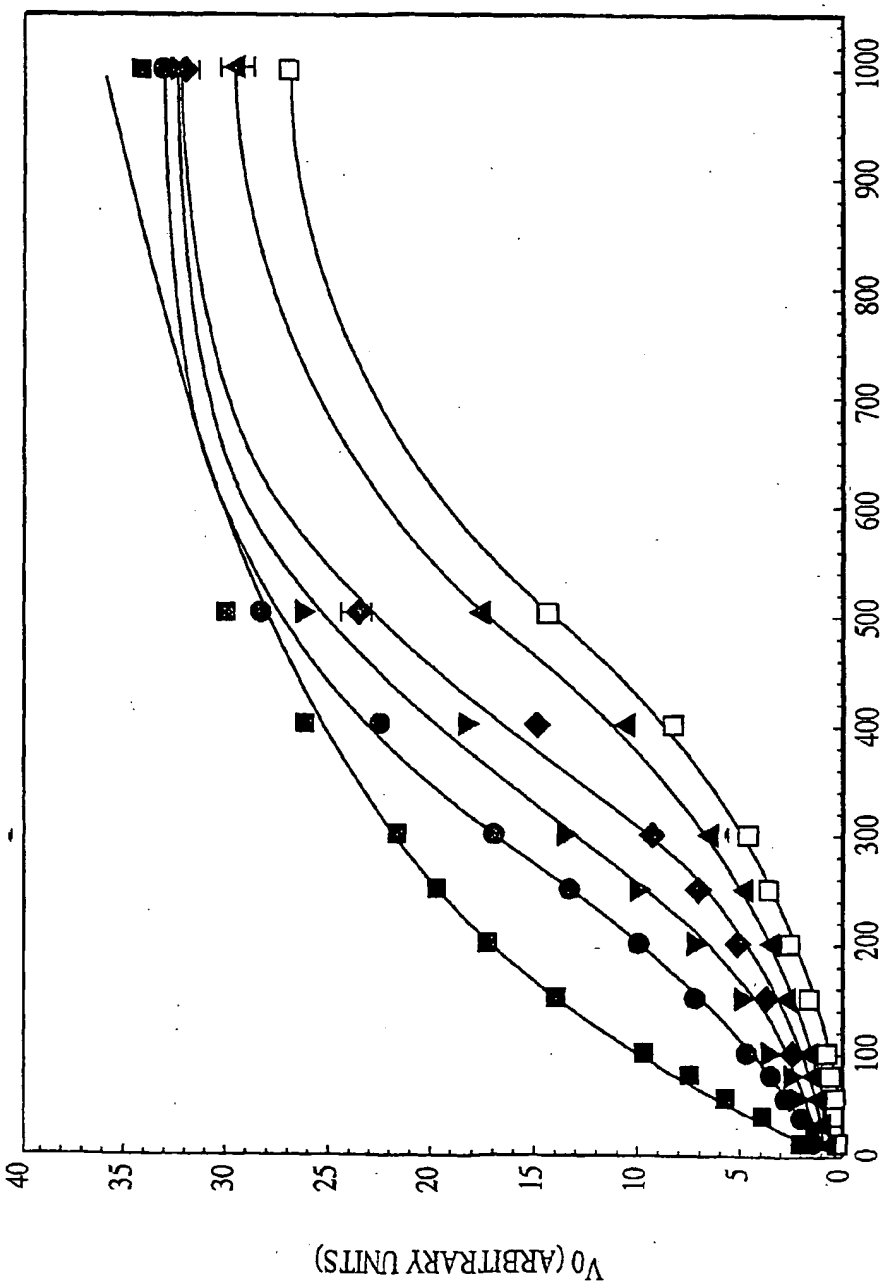
V_0 (ARBITRARY UNITS)
0
5
10
15
20
25
30
35
40
0
100
200
300
400
500
600
700
800
900
1000
PROTHROMBIN (nM)

FIG. 3B

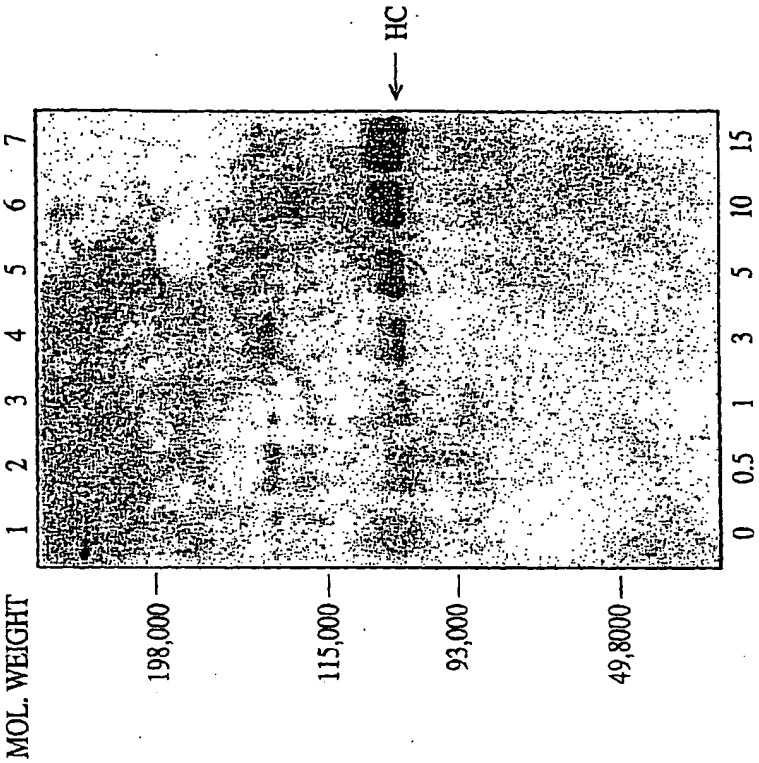
HC
MOL. WEIGHT
198,000
115,000
93,000
49,8000
0 0.5 1 2 3 4 5 6 7
1 3 5 10 15
MIN

FIG. 4B

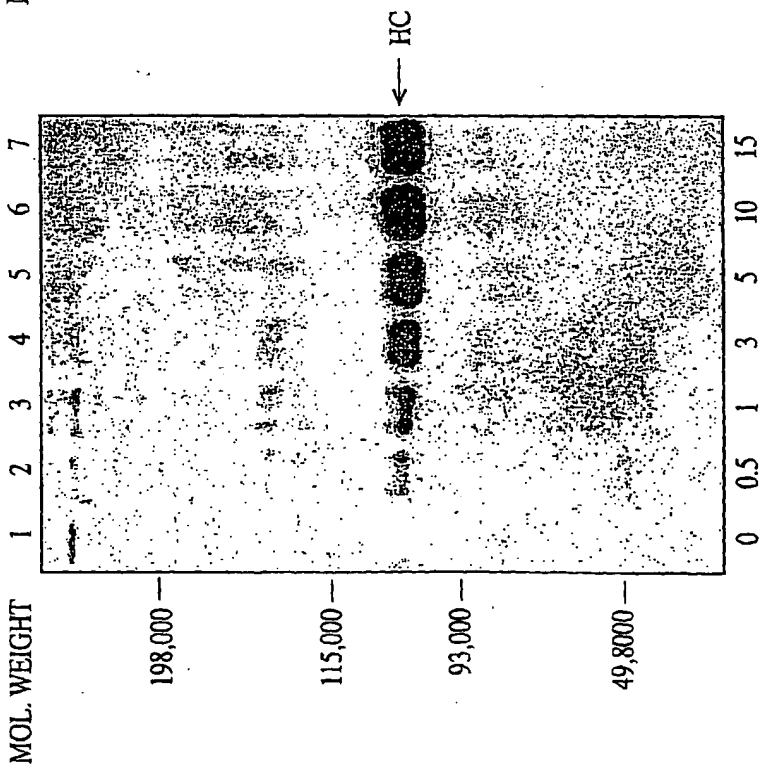
HC
MOL. WEIGHT
198,000
115,000
93,000
49,8000
0 0.5 1 2 3 4 5 6 7
1 3 5 10 15
MIN

FIG. 4A

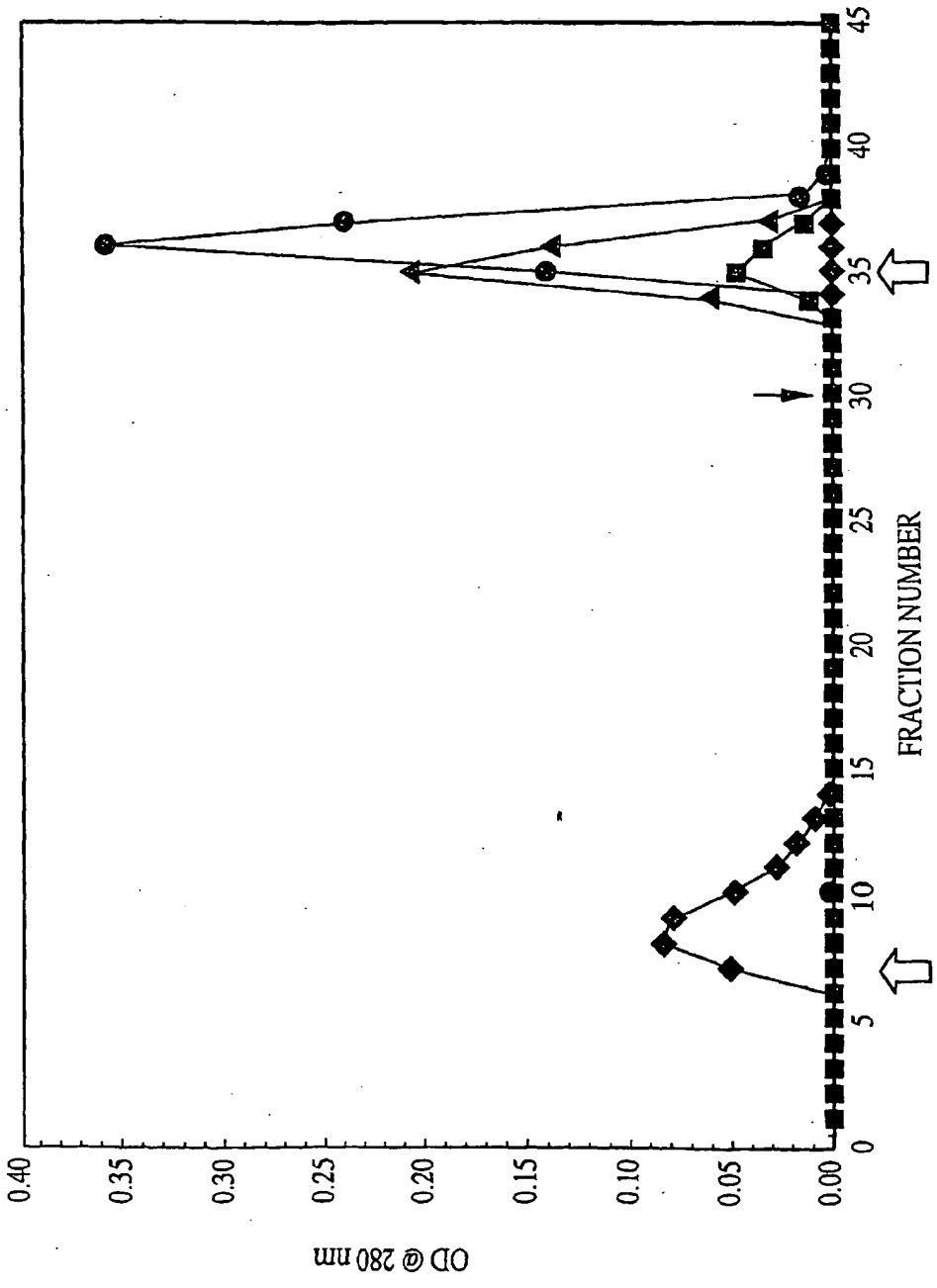
OD @ 280 nm
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
0
5
10
15
20
25
30
35
40
45
FRACTION NUMBER

FIG. 5

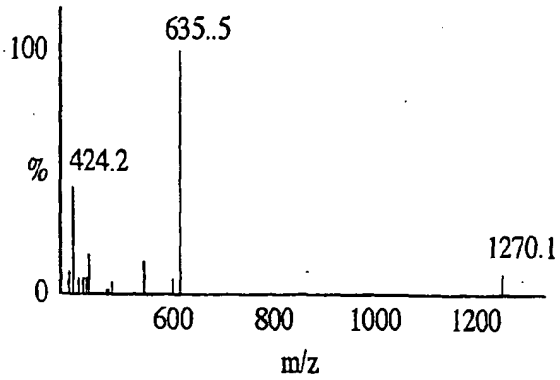

FIG. 5A

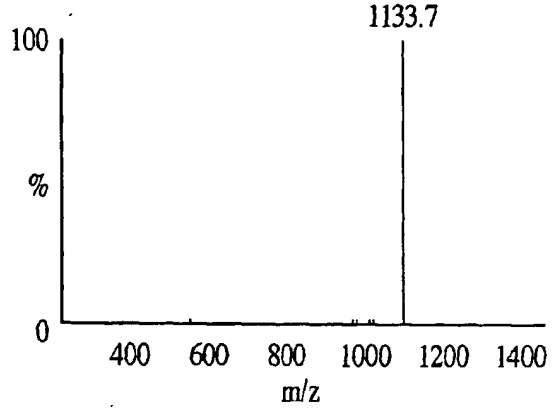

FIG. 5B

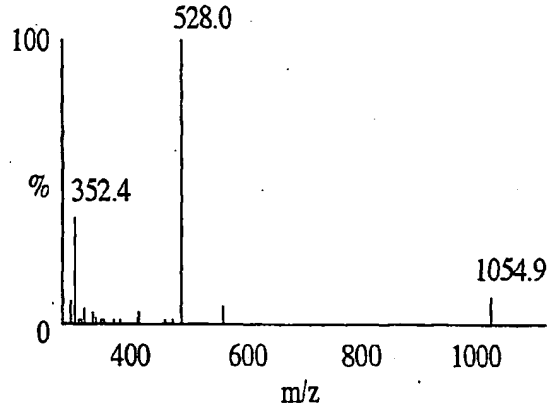

FIG. 5C

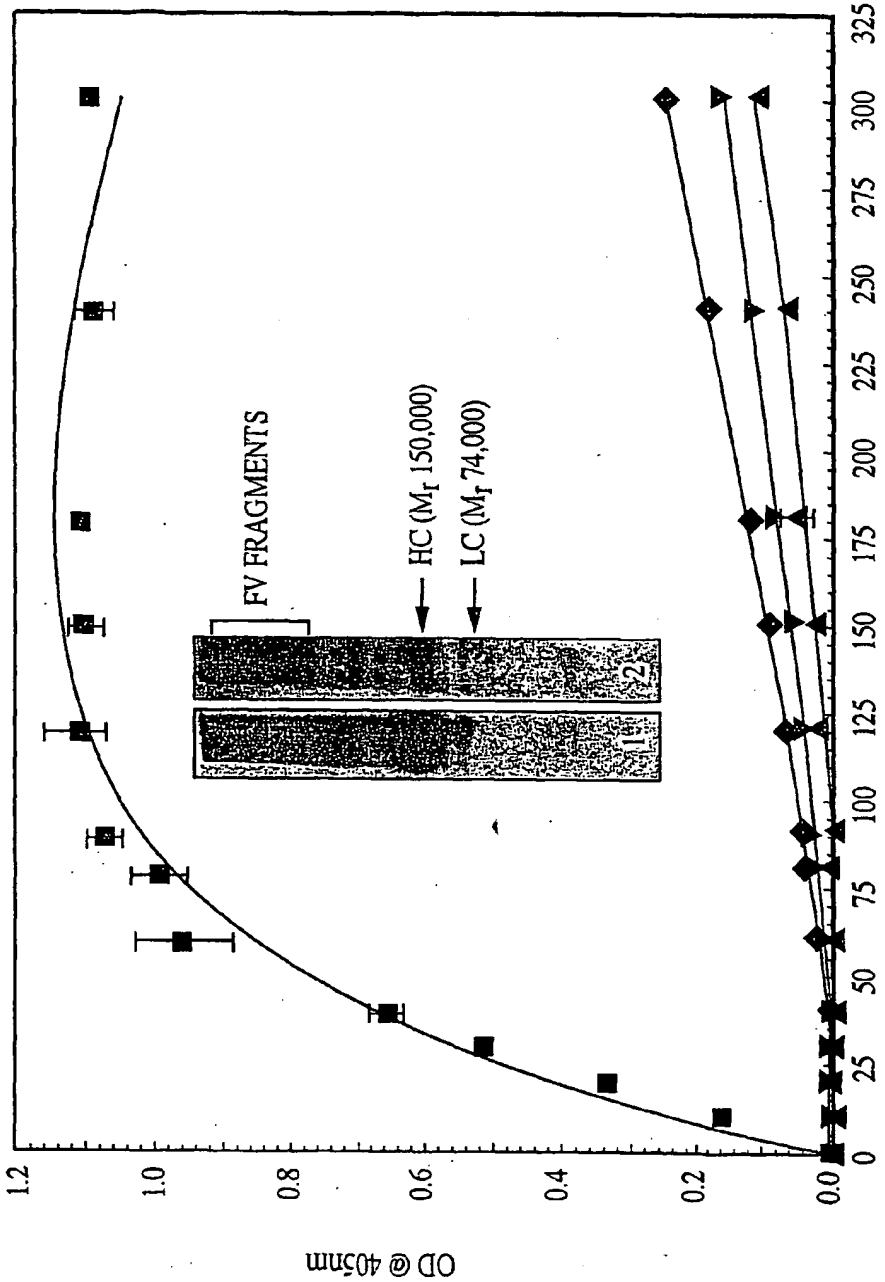
OD @ 405nm
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0
25
50
75
100
125
150
175
200
225
250
275
300
325
SECONDS
FV FRAGMENTS
HC (M_r 150,000)
LC (M_r 74,000)
1
2

FIG. 6A

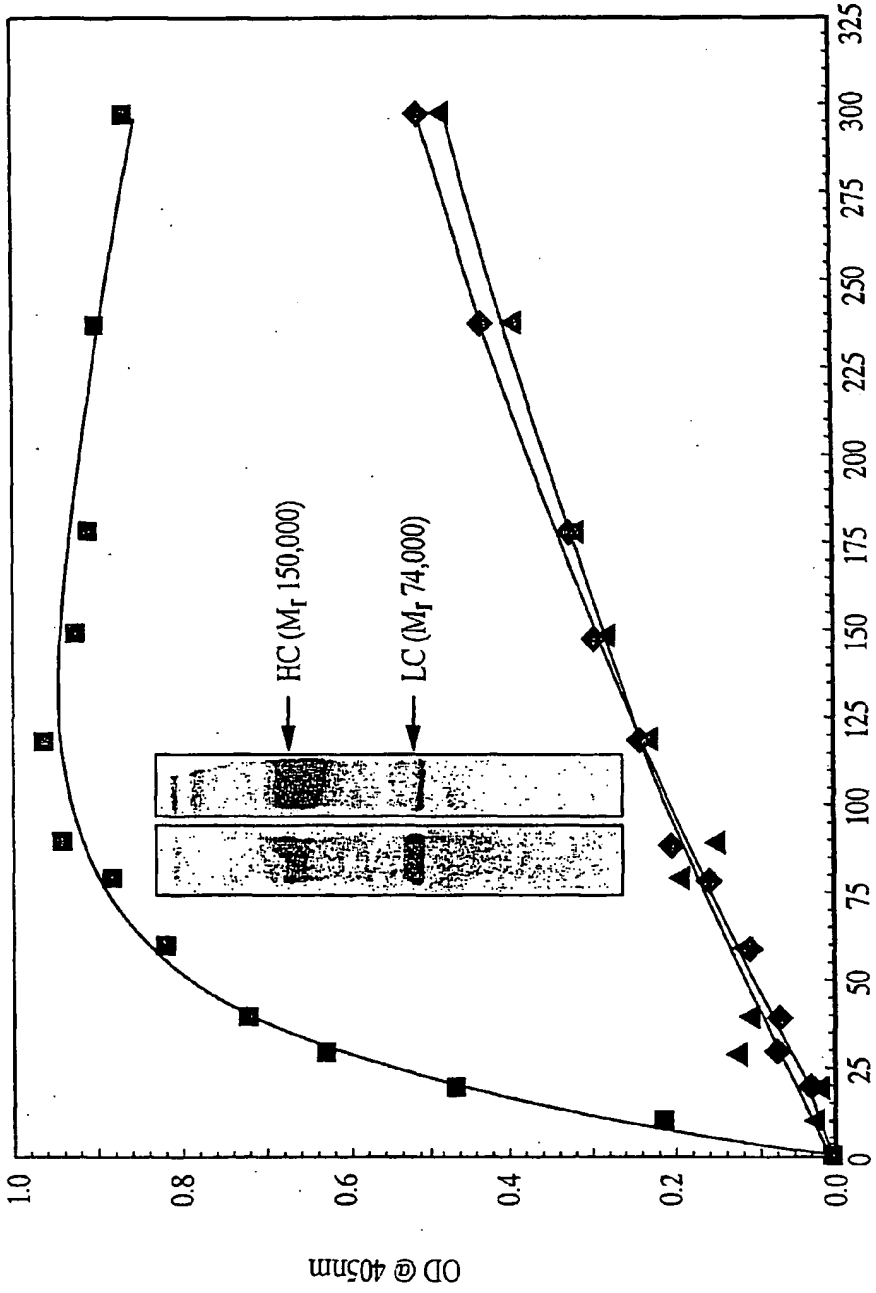
OD @ 405nm
1.0
0.8
0.6
0.4
0.2
0.0
0
25
50
75
100
125
150
175
200
225
250
275
300
325
SECONDS
HC (M_r 150,000)
LC (M_r 74,000)

FIG. 6B

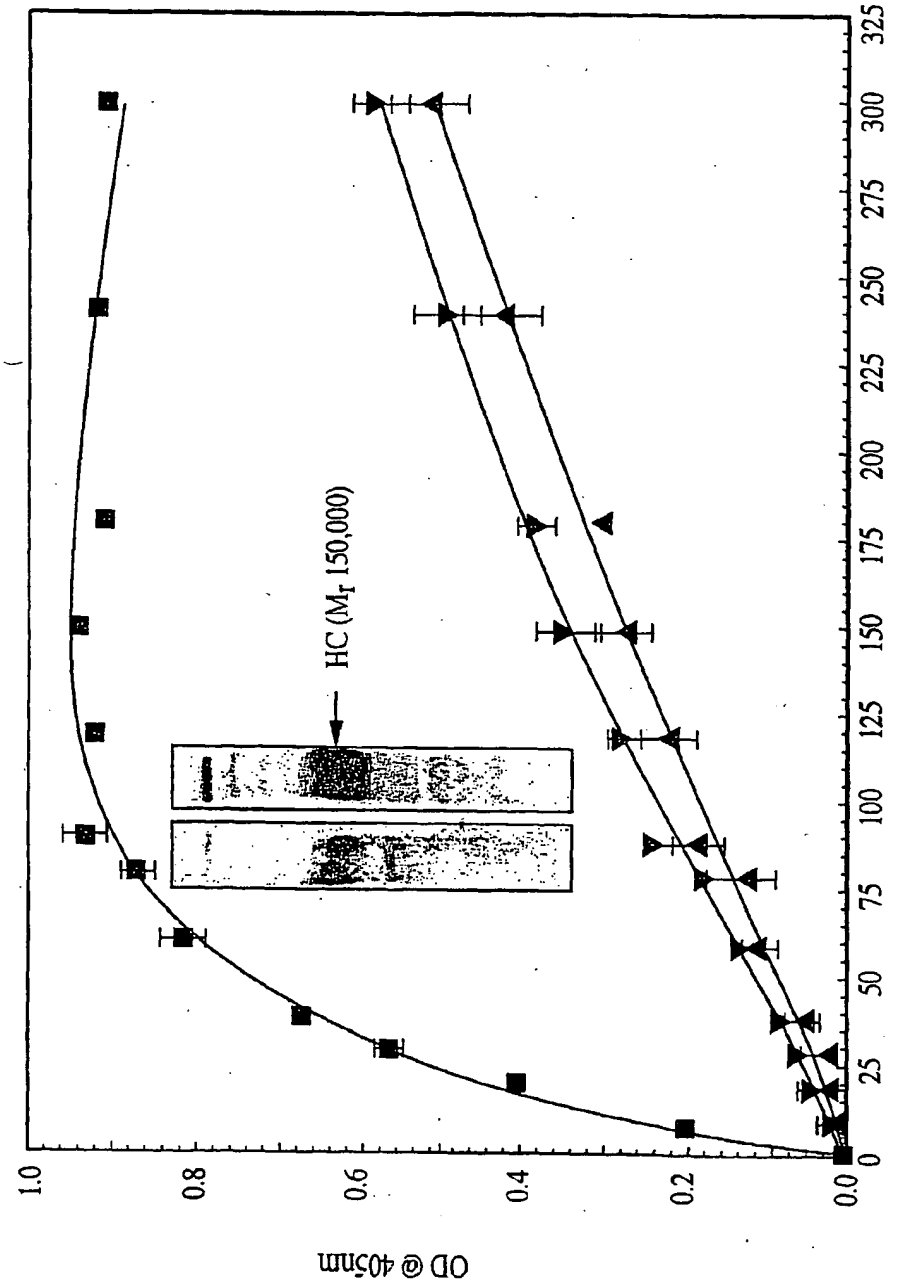
OD @ 405nm
1.0
0.8
0.6
0.4
0.2
0.0
0
25
50
75
100
125
150
175
200
225
250
275
300
325
SECONDS
HC (M_r 150,000)

FIG. 6C

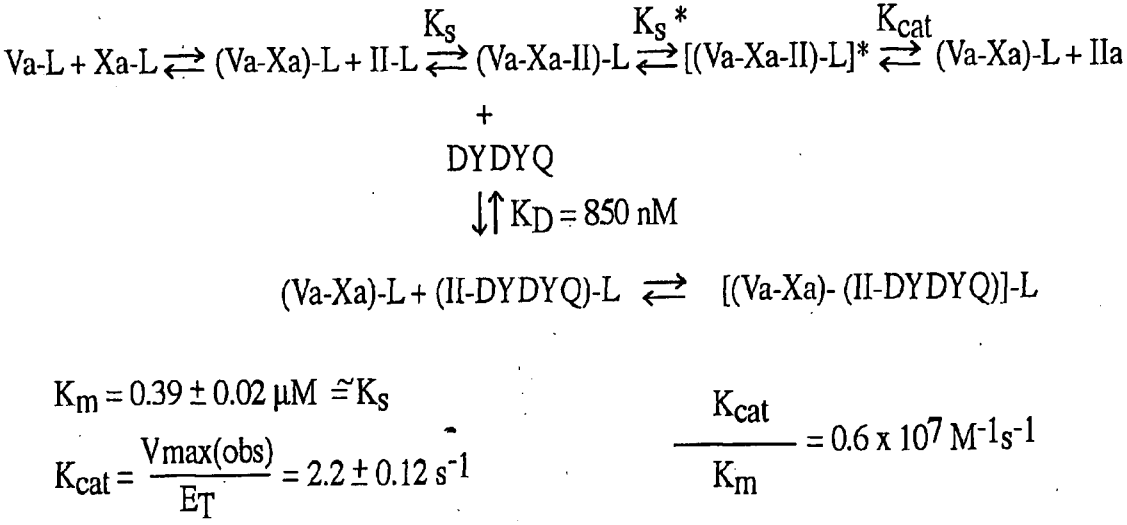
Va-L + Xa-L ⇄ (Va-Xa)-L + II-L ⇄ (Va-Xa-II)-L ⇄ [(Va-Xa-II)-L]* ⇄ (Va-Xa)-L + IIa
Ks
Ks *
Kcat
+
DYDYQ
↓↑ KD = 850 nM
(Va-Xa)-L + (II-DYDYQ)-L ⇄ [(Va-Xa)- (II-DYDYQ)]-L
Km = 0.39 ± 0.02 μM ≅ Ks
Kcat = Vmax(obs) / ET = 2.2 ± 0.12 s-1
Kcat / Km = 0.6 x 10^7 M-1s-1

FIG. 7


DY1DYQ
SO3 2-
(D5Q1)
SEQ ID NO. 7
DYDY2Q
SO3 2-
(D5Q2)
SEQ ID NO. 8
DY1DY2Q
SO3 2-
SO3 2-
(D5Q1,2)
SEQ ID NO. 9

FIG. 8

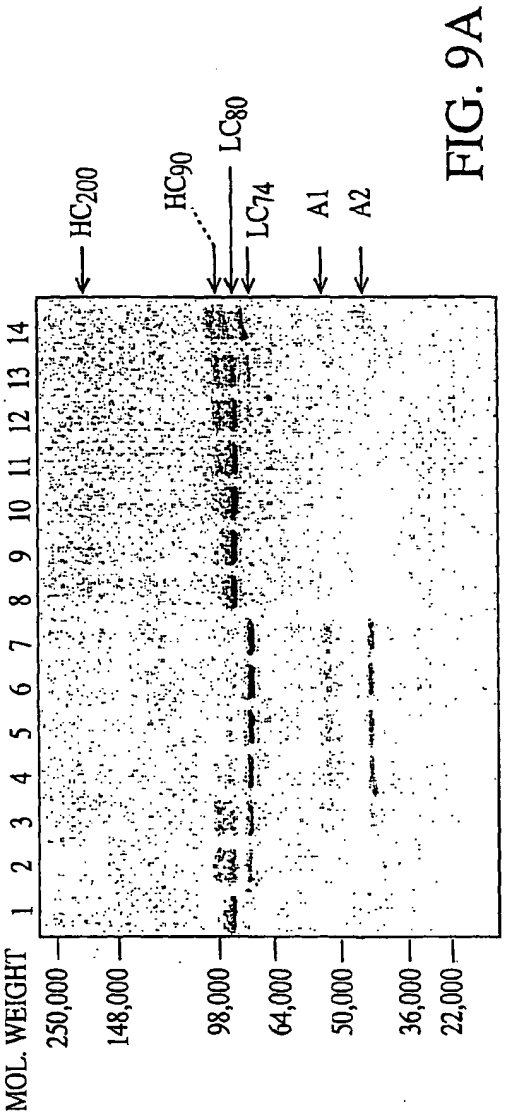
MOL. WEIGHT
250,000
148,000
98,000
64,000
50,000
36,000
22,000
1 2 3 4 5 6 7 8 9 10 11 12 13 14
HC200
HC90
LC80
LC74
A1
A2

FIG. 9A

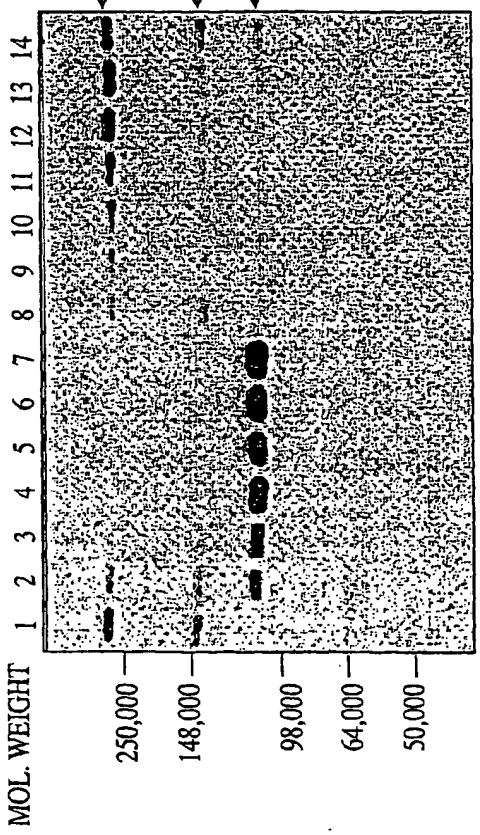
MOL. WEIGHT
250,000
148,000
98,000
64,000
50,000
1 2 3 4 5 6 7 8 9 10 11 12 13 14
FV
HC

FIG. 9B

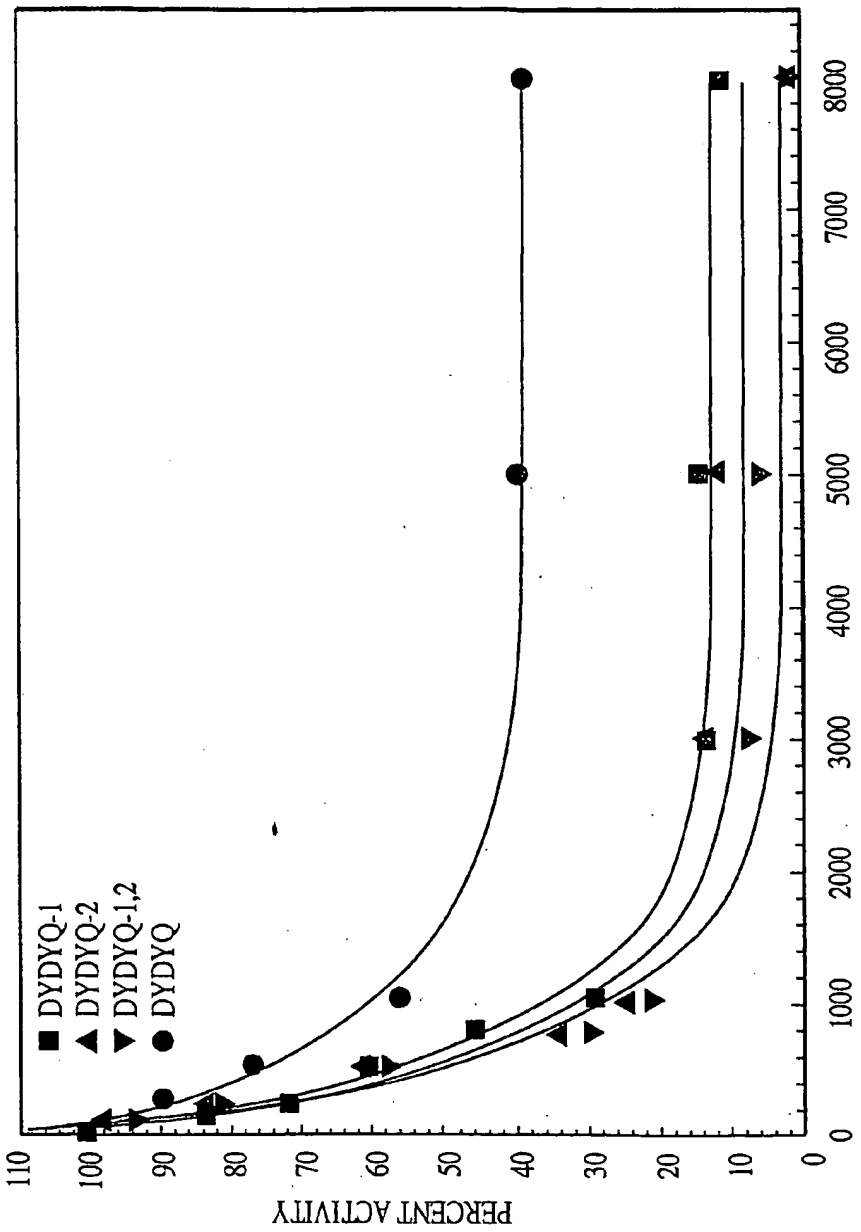
DYDYQ-1
DYDYQ-2
DYDYQ-1,2
DYDYQ
PERCENT ACTIVITY
110
100
90
80
70
60
50
40
30
20
10
0
0
1000
2000
3000
4000
5000
6000
7000
8000
(DYDYQ1,2 nM)

FIG. 10A

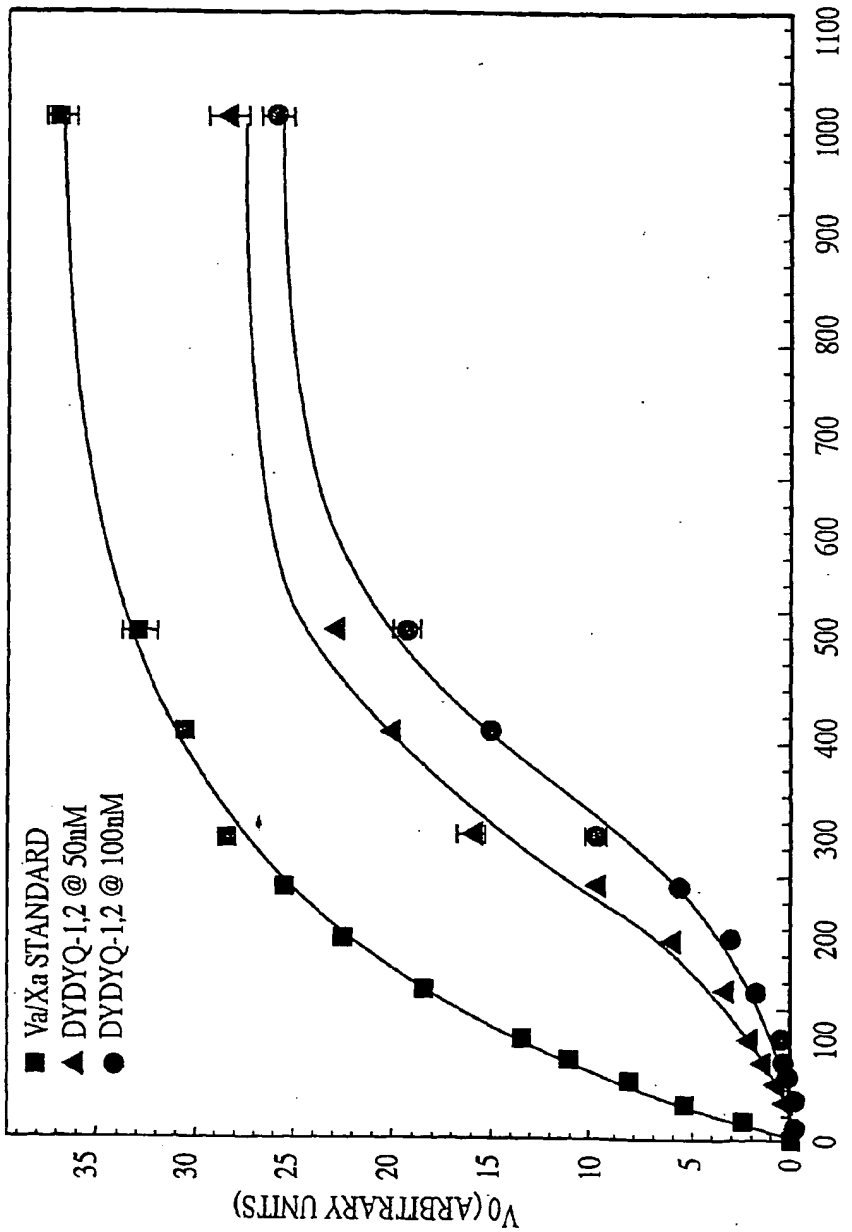
Va/Xa STANDARD
DYDYQ-1,2 @ 50nM
DYDYQ-1,2 @ 100nM
V0 (ARBITRARY UNITS)
PROTHROMBIN (nM)
0
5
10
15
20
25
30
35
0
100
200
300
400
500
600
700
800
900
1000
1100

FIG. 10B

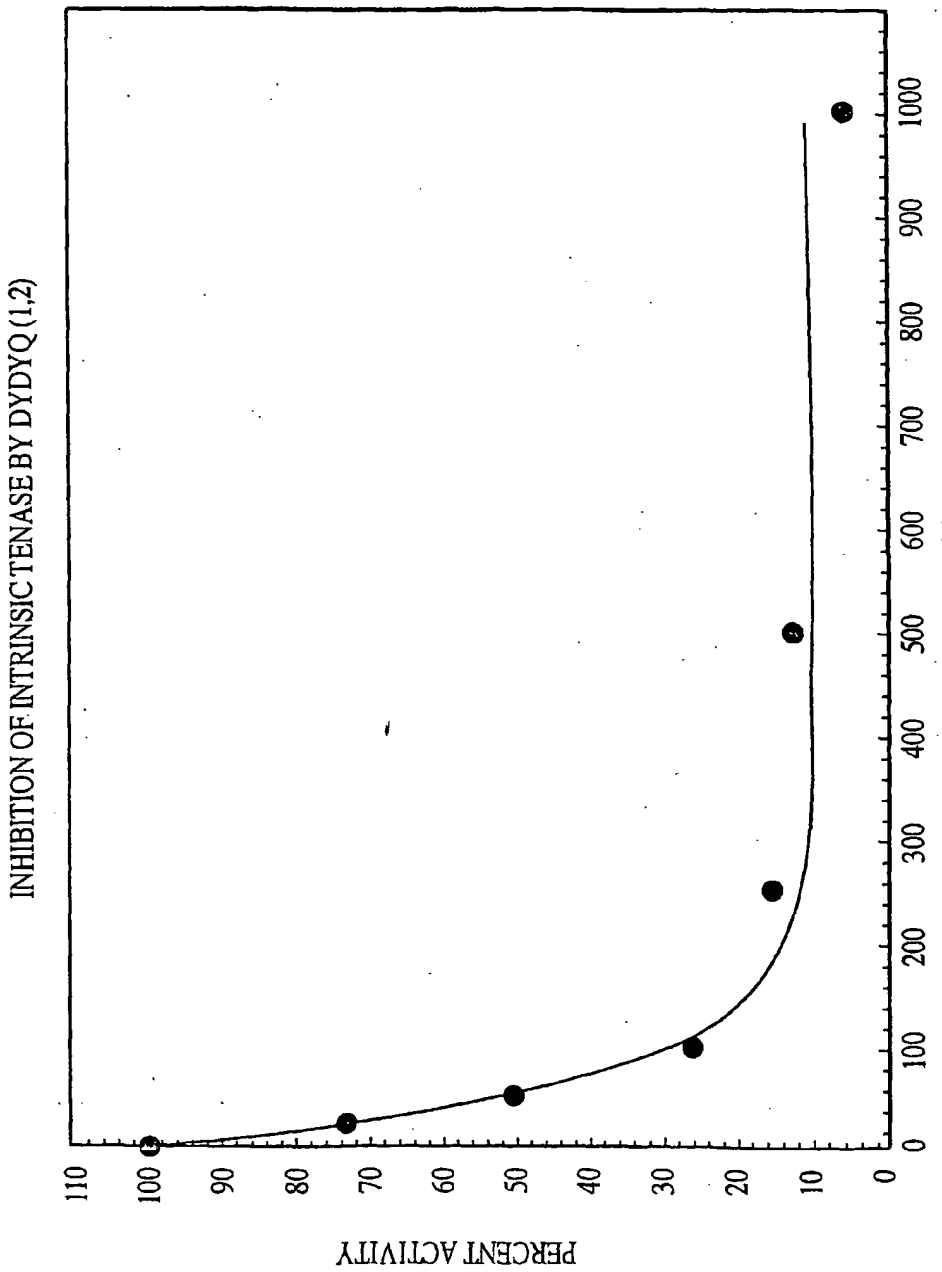
INHIBITION OF INTRINSIC TENASE BY DYDYQ (1,2)
PERCENT ACTIVITY
110
100
90
80
70
60
50
40
30
20
10
0
0
100
200
300
400
500
600
700
800
900
1000
(DYDYQ1,2 nM)

FIG. 11A

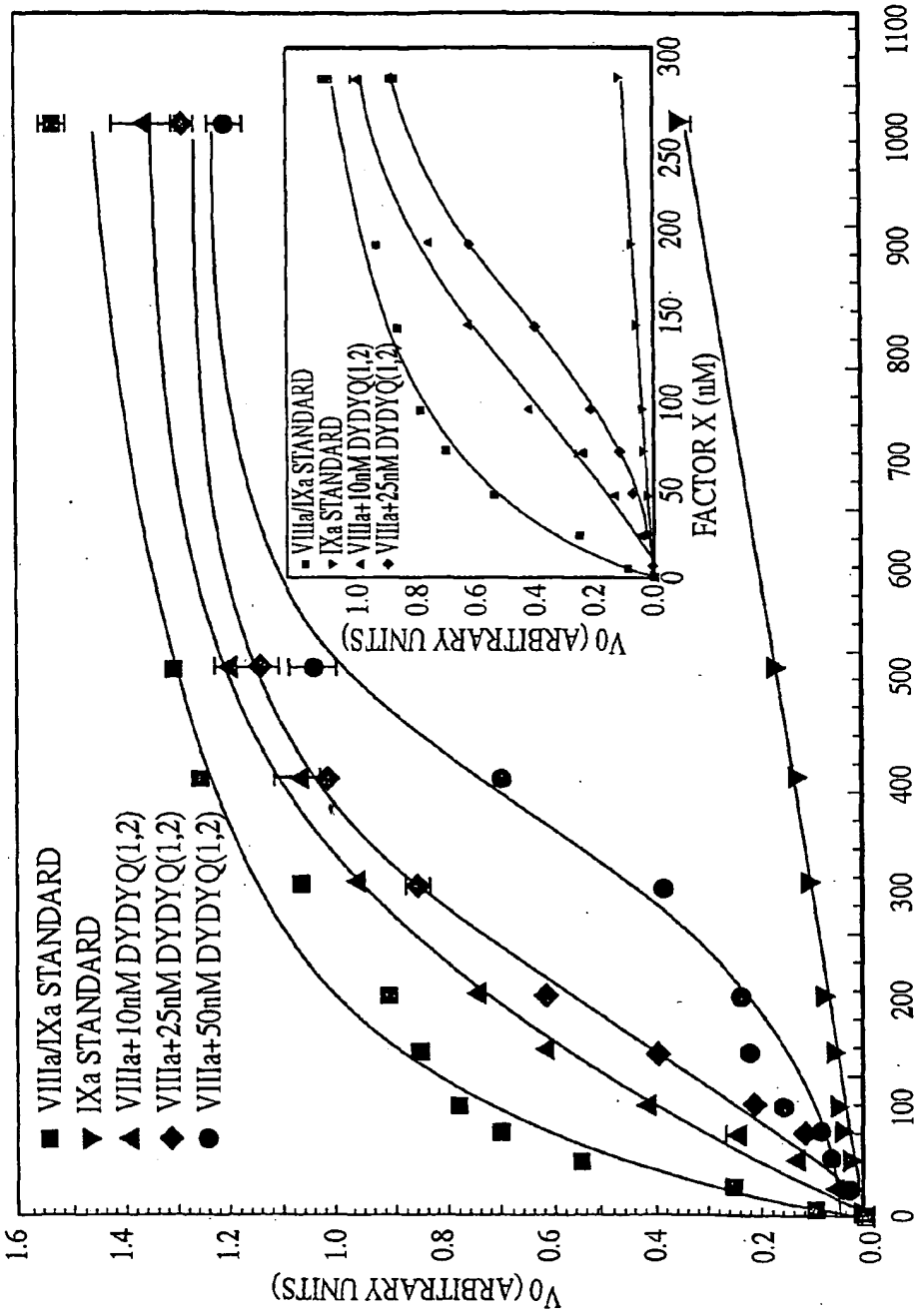
VIIIa/IXa STANDARD
IXa STANDARD
VIIIa+10nM DYDYQ(1,2)
VIIIa+25nM DYDYQ(1,2)
VIIIa+50nM DYDYQ(1,2)
V0 (ARBITRARY UNITS)
FACTOR X (nM)
0.0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
1.6
0
100
200
300
400
500
600
700
800
900
1000
1100
VIIIa/IXa STANDARD
IXa STANDARD
VIIIa+10nM DYDYQ(1,2)
VIIIa+25nM DYDYQ(1,2)
V0 (ARBITRARY UNITS)
FACTOR X (nM)
0.0
0.2
0.4
0.6
0.8
1.0
0
50
100
150
200
250
300

FIG. 11B

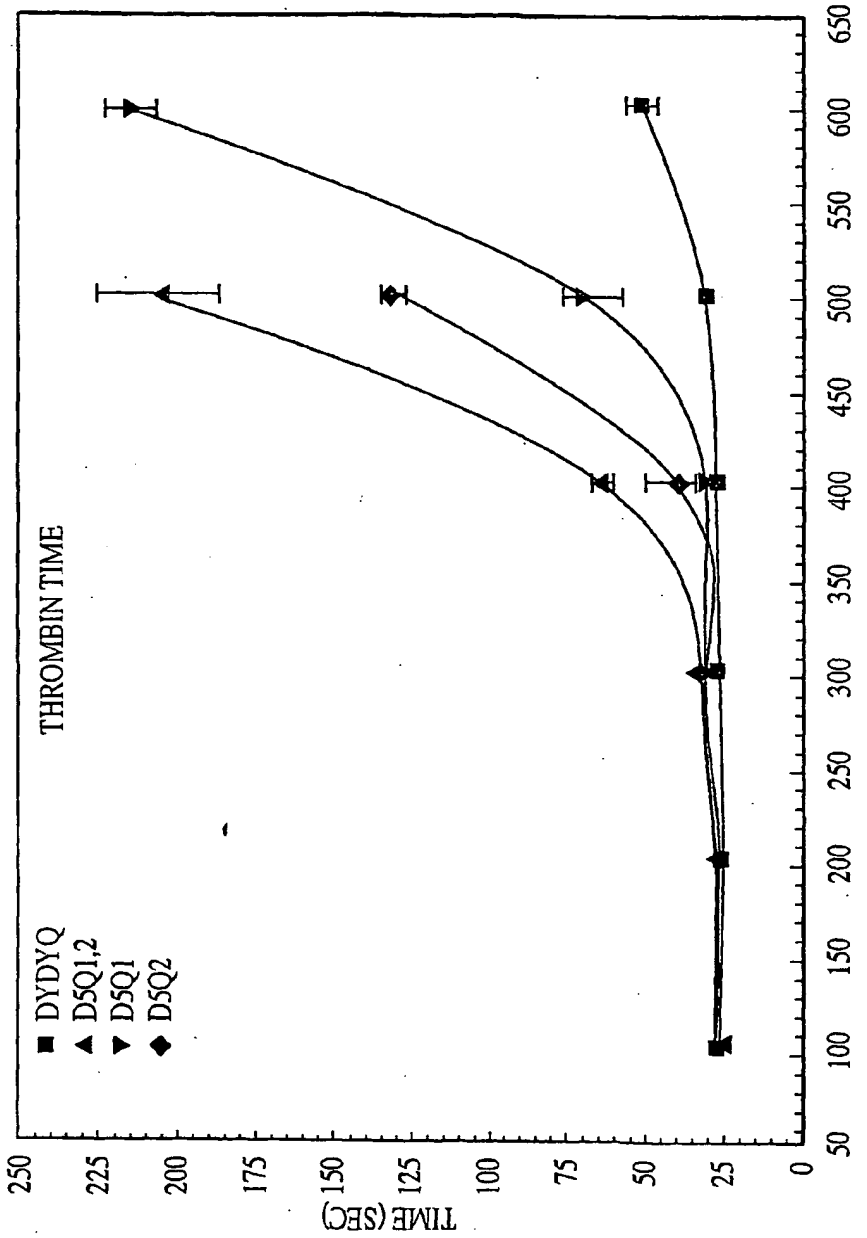
THROMBIN TIME
DYDYQ
D5Q1,2
D5Q1
D5Q2
TIME (SEC)
PEPTIDE (μM)
0
25
50
75
100
125
150
175
200
225
250
50
100
150
200
250
300
350
400
450
500
550
600
650

FIG. 12

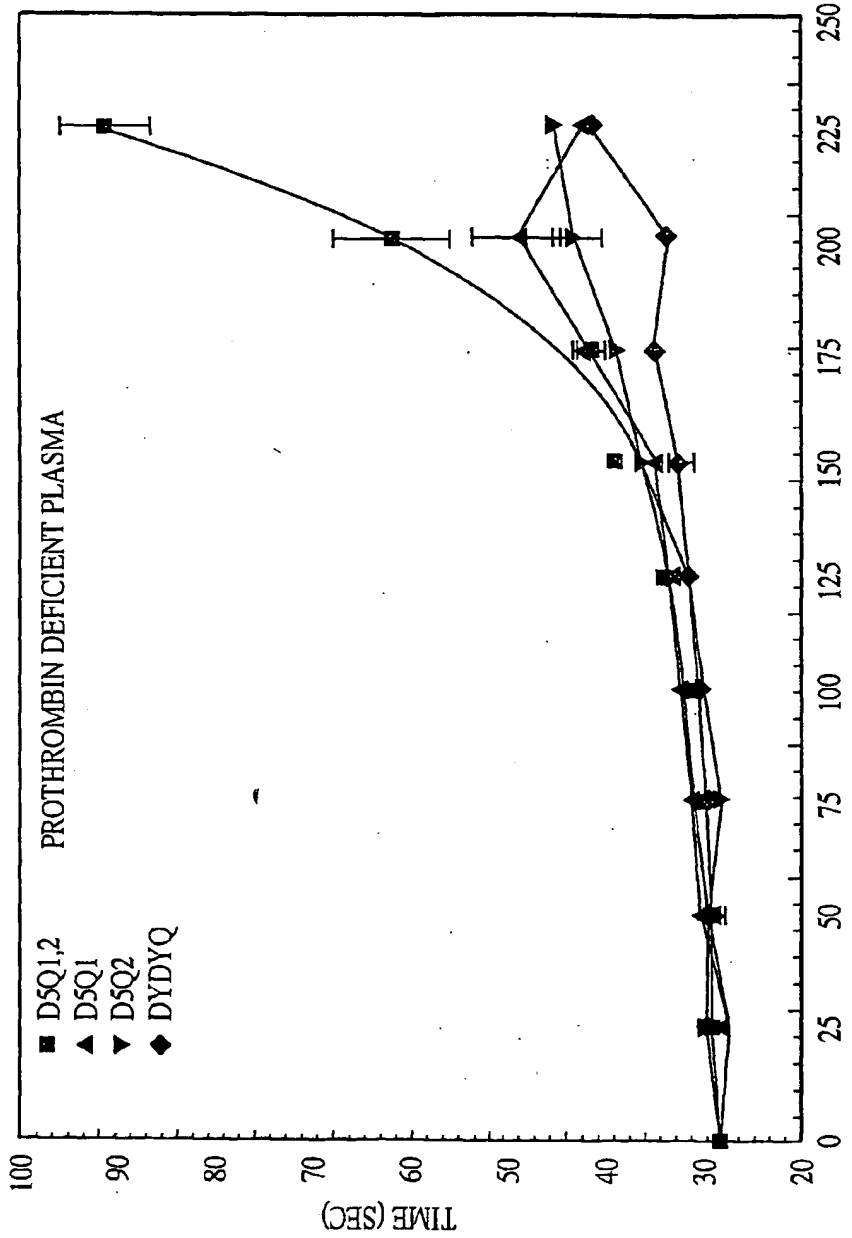
PROTHROMBIN DEFICIENT PLASMA
D5Q1,2
D5Q1
D5Q2
DYDYQ
TIME (SEC)
PEPTIDE (μM)
100
90
80
70
60
50
40
30
20
0
25
50
75
100
125
150
175
200
225
250

FIG. 13